

# The investigation of the sludge reduction efficiency and mechanisms in oxic–settling–anaerobic (OSA) process

Özlem Demir and Ayşe Filibeli

## ABSTRACT

This paper aims to provide a full understanding of the sludge reduction mechanisms in the oxic–settling–anaerobic (OSA) process and presents an evaluation of the sludge reduction efficiencies and sludge characteristics in this process compared to the conventional activated sludge process. Fifty-eight percent reduction in observed yield in the OSA process was achieved compared to the control system at the end of the operational period with no deterioration of effluent quality. The settleability of sludge in the OSA process was also found to be better than that of the control system in terms of sludge volume index. In long-term operation, capillary suction time and specific resistance to filtration values confirmed that the OSA process showed good filterability characteristics. The results of batch experiments showed that higher endogenous respiration in the systems might lead to lower sludge production and that energy uncoupling had only a limited impact on sludge reduction.

**Key words** | cell lysis, energy uncoupling, OSA (oxic–settling–anaerobic) process, sludge minimization during biological treatment, sludge reduction technologies

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## INTRODUCTION

Biological wastewater treatments transform dissolved and suspended organic contaminants into biomass and evolved gases. The most widely used biological wastewater treatment method in the world for both domestic and industrial wastewater is the activated sludge process. However, there are some problems related to the application of this processes. One of the main problems of the conventional activated sludge processes is high sludge production. This creates major economic, environmental and legal challenges for biological treatment plants as they struggle to dispose of excess sludge (Wei *et al.* 2003; Ødegaard 2004). The amount of sludge remaining after treatment is still so high that significant costs are incurred for the treatment and disposal of excess sludge in a biological wastewater treatment system. In addition to this, the main options for sludge treatment and disposal such as landfill, incineration and agricultural reuse have some disadvantages such as the production of gaseous emissions and hazardous compounds. Agricultural reuse of sludge is often strictly regulated due to the potential health risks related to pathogens and contaminants such as heavy metals. Incineration is another option which remains cost

intensive and leads to the loss of organic matter and nutrients (Foladori *et al.* 2010). Bearing all of these issues in mind, sludge minimization techniques become more favorable options than conventional sludge treatment methods (Semblante *et al.* 2014). One of the ways to overcome sludge problems is to reduce sludge production in the wastewater treatment itself, rather than relying on post-treatment. Wastewater treatment must encourage non-growth activities through biosynthesis in order to reduce biomass production (Wei *et al.* 2003; Ødegaard 2004). Sludge reduction technologies are based on different strategies including lysis–cryptic growth, uncoupling metabolism and micro-fauna predation (Egemen *et al.* 2001; Chen *et al.* 2002; Saby *et al.* 2002; Wei *et al.* 2003; He *et al.* 2006; Liang *et al.* 2006; Wei & Liu 2006; Guo *et al.* 2007; Li *et al.* 2008).

Among these sludge reduction technologies, the oxic–settling–anaerobic (OSA) process is a promising option using an anaerobic sludge tank in the return sludge line of conventional activated sludge process (Zhou *et al.* 2015). The OSA process at laboratory scale was used to evaluate the reduction of excess sludge for the first time by Chudoba *et al.* (1992). In the OSA process, a part of

the return activated sludge is treated in an anaerobic tank at low oxidation reduction potential (ORP) level and then returned to the aerobic reactor (Semblante *et al.* 2015). The OSA process presents several advantages such as being easily implemented as part of existing conventional activated sludge systems (CAS), bringing about sludge reduction and improvement of sludge settleability without effluent quality deterioration (Pérez-Elvira *et al.* 2006). The OSA process has delivered significant sludge reduction efficiency without negative effects on pollutant removal efficiency and sludge settleability (Zhou *et al.* 2015).

The possible sludge reduction mechanisms in the OSA process have been investigated by several researchers. Perez Elvira *et al.* (2006) reported that alternate anaerobic–aerobic cycling of activated sludge could stimulate catabolic activity. Thus, the dissociation of catabolism from anabolism leads to sludge reduction in the OSA process. The aerobic microorganisms use energy produced from organic matter oxidation in the aerobic tank but these microorganisms cannot produce the required energy under the anaerobic, food-limited condition and therefore, they use their stored energy. When they return to the aeration tank, they rebuild adenosine triphosphate (ATP) reserves before the new cell synthesis, leading to sludge reduction (Khursheed *et al.* 2015). Another hypothesis for the explanation of sludge reduction in the OSA process is the disruption of the cell wall, resulting in cell lysis (Chen *et al.* 2003; Saby *et al.* 2003). According to this hypothesis, the anaerobic reactor serves as a cell lysis reactor. When the sludge returns to the aerobic tank, the soluble chemical oxygen demand (SCOD) released from cell lysis leads to cryptic growth and sludge reduction (Foladori *et al.* 2010).

Both sludge treatment and minimization techniques were evaluated in the literature detailed. However, the studies related to sludge reduction by the OSA process are limited. In this study, a laboratory-scale OSA process, a modification of CAS using an anaerobic tank prior to an aerobic tank, was established. A CAS system as control system was also operated in parallel under the same conditions to allow for a comparison and evaluation of the sludge reduction. As a further investigation of the sludge reduction mechanisms in the OSA process, batch experiments were conducted. Briefly, this paper aims to provide a full understanding of the sludge reduction mechanism in the OSA process and present an evaluation of sludge reduction efficiency and sludge characteristic in this process compared to the CAS.

## MATERIAL AND METHODS

### System operation

Two identical laboratory-scale, extended aeration activated sludge systems were designed and operated simultaneously. The volumes of the oxic reactors and settlers were 6 L and 3 L, respectively, in both laboratory-scale reactors. Bubble diffusers located at the bottom of the oxic reactors maintained 2–3 mg/L dissolved oxygen (DO) concentration. In both systems, the hydraulic retention times and solid retention times were 25 h and 25 d, respectively. The return sludge flow rate was 0.75 for the systems. The seed, which is characterized in Table 1, was obtained from the aeration tank of the Kemalpaşa Pakmaya Treatment Plant in İzmir for the start-up of the systems.

Synthetic wastewater composed of the different compounds listed in Table 2 was fed into the systems continuously with a fixed chemical oxygen demand (COD) strength of 400 mg/L. The concentrations of the components were proportionally adjusted to maintain constant C/N/P =

**Table 1** | The properties of activated sludge from Pakmaya Treatment Plant

Parameter	Average value
pH	7.26
Temperature (°C)	27.8
ORP (mV)	68
SVI (mL/g)	123
MLSS (mg/L)	6,325
MLVSS (mg/L)	2,675
Total solids (mg/L)	11,205
Volatile solids (mg/L)	3,175
NH <sub>4</sub> -N (mg/L)	58
PO <sub>4</sub> -P (mg/L)	51
NO <sub>3</sub> -N (mg/L)	128
CST (s)	69

**Table 2** | Synthetic wastewater composition

Component	Concentration (g/L)
Molasses (mL)	48
NH <sub>4</sub> Cl (g/L)	0.076
KH <sub>2</sub> PO <sub>4</sub> (g/L)	0.0176
MgSO <sub>4</sub> ·7H <sub>2</sub> O (g/L)	0.0225
CaCl <sub>2</sub> (g/L)	0.05426
FeCl <sub>3</sub> (g/L)	0.00025

100/5/1 ratio. The ORP in the sludge holding tank used in the OSA process was maintained at  $-250$  mV.

### Modification of activated sludge process as OSA process

Before the modification, two CAS systems were operated simultaneously for a 45-day period to achieve steady state conditions. After reaching stabilized conditions, one of the systems was left unmodified as a control system, Figure 1(a); the other system was modified by inserting a 3 L anaerobic tank into the sludge recycling line to form the OSA process depicted in Figure 1(b). In the OSA process, 20% of return activated sludge was subjected to a low ORP level and food-limited conditions in the anaerobic tank. Pure nitrogen was injected manually to maintain a low ORP level. The control system and OSA process were also operated over the course of 45 days to monitor effluent quality, sludge production and sludge characteristics. The possible mechanisms responsible for sludge reduction in the OSA process were also investigated. Two batch experiments for the endogenous decay of sludge and uncoupling metabolism were carried out in order to identify the sludge reduction mechanisms operating in the OSA process.

### Investigation of sludge reduction mechanisms

#### Batch experiment I: sludge decay

In order to investigate the sludge decay theory, two reactors were used for batch experiments. Each reactor had 2 L

volume. One litre of sludge taken from the aeration tank of the control system and 1 L of sludge from the OSA process were transferred to the batch reactors. The batch reactors were operated with a 24 h cycle: 6 h aerobic reaction and 18 h anaerobic reaction for sludge at  $-250$  mV of low ORP level. The aim of this batch experiment was to investigate the relationship between sludge anaerobic reaction time, sludge lysis and sludge yield. At the beginning and at the end of the anaerobic cycle, mixed liquor suspended solids (MLSS), COD,  $\text{PO}_4\text{-P}$ ,  $\text{NH}_4\text{-N}$  and total phosphorus (TP) concentrations and oxygen uptake rate (OUR) were monitored.

#### Batch experiment II: energy uncoupling

In order to investigate the energy uncoupling theory, a batch test was conducted. According to the energy uncoupling theory (Chen *et al.* 2003) sludge yield will decline more during exposure to a food-sufficient oxic condition compared to a food-insufficient anaerobic condition. One litre of sludge sample was taken from the aeration tank of the control system and then aerated for 7 h. At the beginning and at the end of the aerobic treatment, MLSS and COD concentrations were measured. In addition to this, 1 L of sludge sample taken from the aeration tank of the control system was subjected to anaerobic conditions under  $-250$  mV of ORP level for 7 h via pure nitrogen injection. After the anaerobic cycle, the synthetic wastewater was added to the anaerobic sludge. SCOD of mixed sludge and MLSS concentration were measured at the beginning and at the end of the anaerobic cycle. Sludge yield was calculated based on the changes in MLSS and COD concentrations.

### Analytical methods

COD and  $\text{NH}_4\text{-N}$  removal efficiencies,  $\text{NO}_3\text{-N}$  and  $\text{NO}_2\text{-N}$  concentrations of the effluent,  $\text{PO}_4\text{-P}$  concentrations of influent and effluent, and total nitrogen (TN) and TP concentrations of the aeration tank were also measured for both the OSA process and the control system three times a week.  $Y_{\text{obs}}$  (observed sludge yield) values for the systems were calculated and evaluated. Furthermore, SVI (sludge volume index), SRF (specific resistance to filtration), CST (capillary suction time) and particle size were measured in order to characterize the sludge. OUR and specific oxygen uptake rate (SOUR) were monitored periodically. pH, temperature (T), DO, electrical conductivity (EC) and ORP were monitored daily. In the first batch experiment, at the beginning and at the end of the anaerobic cycle, MLSS, COD,  $\text{PO}_4\text{-P}$ ,  $\text{NH}_4\text{-N}$  and TP concentrations and OUR were determined. ORP level in the anaerobic period

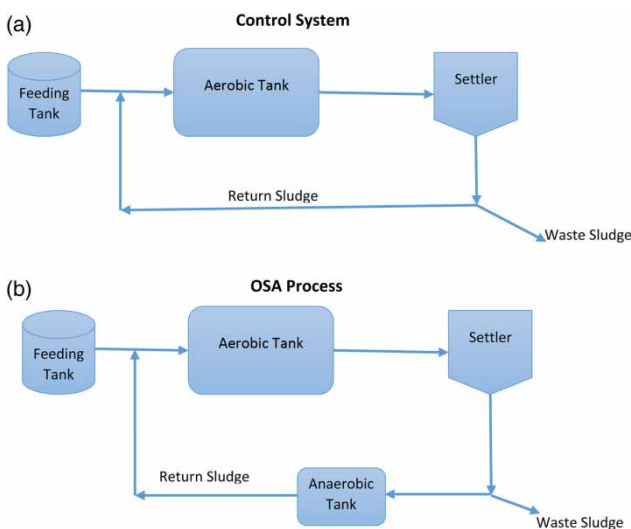


Figure 1 | Schematic experimental setup. (a) Control system, (b) OSA process.

was observed continuously. For the second batch experiment, MLSS, COD, SCOD, and mixed liquor volatile suspended solids (MLVSS) concentrations were analyzed and ORP levels were monitored.

WTW model 340i multi analyzer was used for the measurement of pH, T, DO, EC and ORP. COD concentration was analyzed according to *Standard Methods* (Rice *et al.* 2005). In SCOD measurements, the soluble part of sludge was obtained by centrifuging sludge samples at 10,000 rpm for 20 minutes. The COD solubilization ( $S_{\text{COD}}$ ) was calculated with the following expression, as a percentage (Cui & Jahng 2004; Yan *et al.* 2009):

$$S_{\text{COD}}(\%) = \frac{\text{SCOD}_t - \text{SCOD}_o}{\text{COD}_o - \text{SCOD}_o} \times 100 \quad (1)$$

$\text{SCOD}_o$  = concentration of soluble COD in the untreated sludge (mg/L),

$\text{SCOD}_t$  = concentration of soluble COD in the pre-treated sludge (mg/L),

$\text{COD}_o$  = concentration of total COD in the untreated sludge (mg/L),

MLSS and MLVSS were regularly analyzed according to *Standard Methods* (Rice *et al.* 2005).  $Y_{\text{obs}}$  was calculated as the total solid mass of sludge generated per unit of COD removal.  $\text{NH}_4\text{-N}$  of supernatant was analyzed by using a spectroquant cell test supplied by Merck (Kit number: 14763).  $\text{NO}_3\text{-N}$  of supernatant was analyzed by using a spectroquant cell test supplied by Merck (Kit number: 14773).  $\text{NO}_2\text{-N}$  of supernatant was analyzed by using a spectroquant cell test supplied by Merck (Kit number: 00609). SVI, OUR and SOUR were analyzed according to *Standard Methods* (Rice *et al.* 2005). A Triton 304M CST-meter was used for CST tests. The SRF value of sludge was determined using Buchner funnel test equipment consisting of a graduated cylinder. Particle size distributions were monitored using a Malvern Mastersizer 2000QM.

## RESULTS AND DISCUSSION

### Sludge production in continuous operation of OSA and control system

Compared with the control system,  $Y_{\text{obs}}$  in the OSA process was clearly reduced to a low ORP level as seen in Figure 2(a).

In the OSA process,  $Y_{\text{obs}}$  decreased from 0.52 to 0.2 mgMLSS/mgCOD<sub>removed</sub> during the operation period. Sixty-two percent reduction in yield production was obtained in the OSA process during the operation. In the control system, the average  $Y_{\text{obs}}$  value was 0.52 mgMLSS/mgCOD<sub>removed</sub>. The OSA process also achieved a 58% reduction in  $Y_{\text{obs}}$  compared to the control system at the end of the operation. It can be concluded that a low ORP level in the anaerobic tank promotes excess sludge reduction. In the first study related to the OSA process by Chudoba *et al.* (1992), the sludge production of a CAS system was compared to that of the OSA process at laboratory-scale and using synthetic wastewater. In the case of the OSA process, the anaerobic reactor was maintained at ORP of -250 mV. The observed sludge yield in the activated sludge system was 0.37 kgTSS/kgCOD<sub>removed</sub> (TSS: total suspended solids), while in the OSA process it was 0.22 kgTSS/kgCOD<sub>removed</sub> (a reduction of 40%).

The impact of the anaerobic conditions in the OSA process can be seen in Figure 2(b). The MLSS concentration of the OSA process was around 3,000 mg/L at the beginning of the operation period. When sludge was subjected to low ORP levels in the anaerobic reactor, MLSS concentration reduced by 37% in the OSA process and reached around 1,900 mg/L. However, in the control system, there was no significant change in MLSS concentration during the operational period. The sludge reduction during the OSA process can be attributed to cell death. According to this hypothesis, the anaerobic reactor behaves like a cell lysis reactor and when the sludge returns to the aerobic activated sludge stage, the soluble COD supports cryptic growth, which leads to an overall sludge reduction. MLSS concentration reduced by 37% in the sludge from the OSA process during the treatment. This means that a low ORP and no external food source may promote sludge reduction since such an environment may impose stress upon the microbes. The impact of the anaerobic exposure on the MLVSS can also be seen in Figure 2(c). The MLVSS concentration reduced by 41% in the sludge from the OSA process during the treatment.

The excess sludge production rate of the OSA process at the ORP level of -250 mV in its anaerobic tank was lower than that at the ORP level of +100 to +150 mV. Therefore, it can be concluded that the OSA process can reduce excess sludge effectively and the ORP level in the anaerobic tank may be an important factor for the reduction of excess sludge.

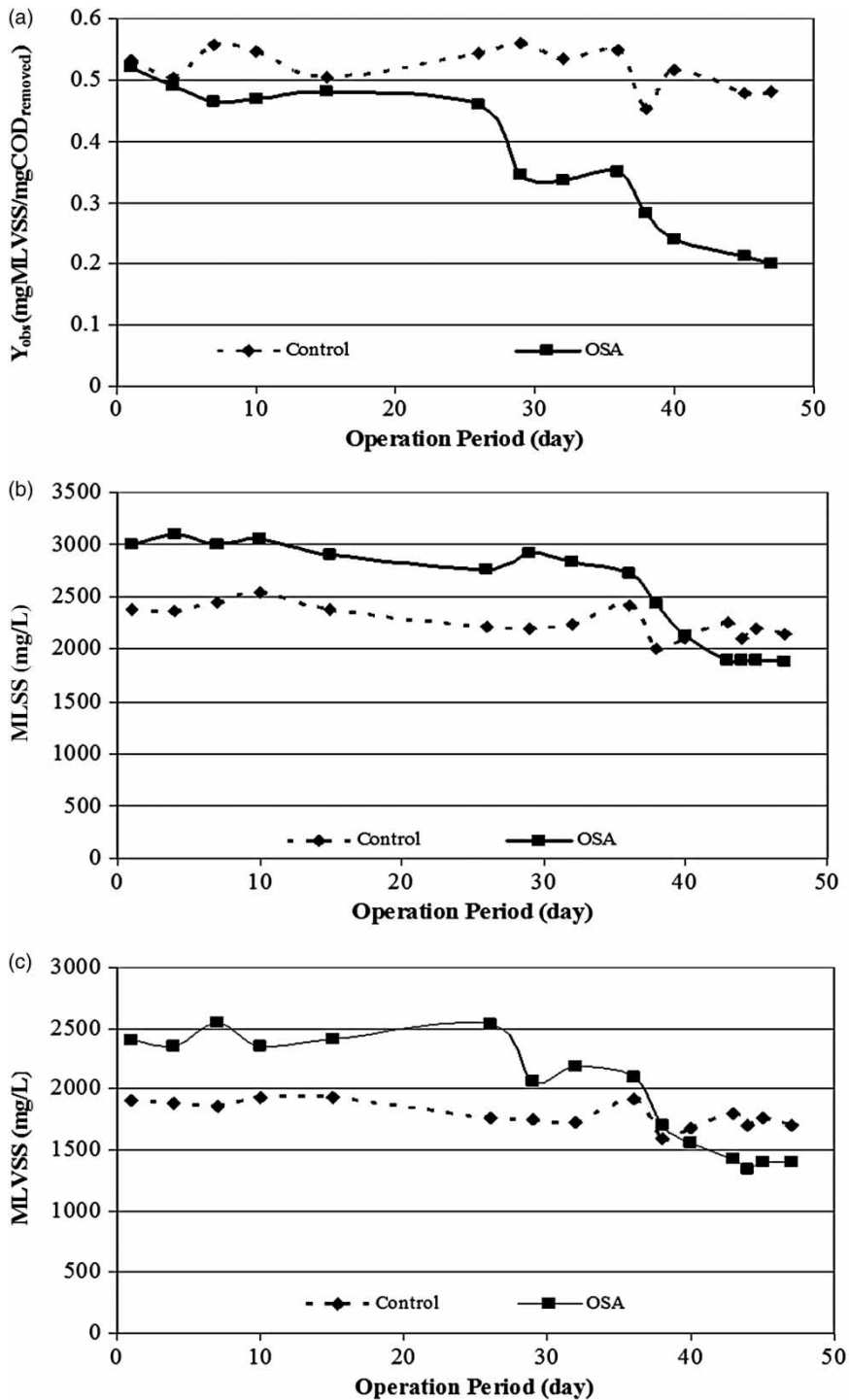
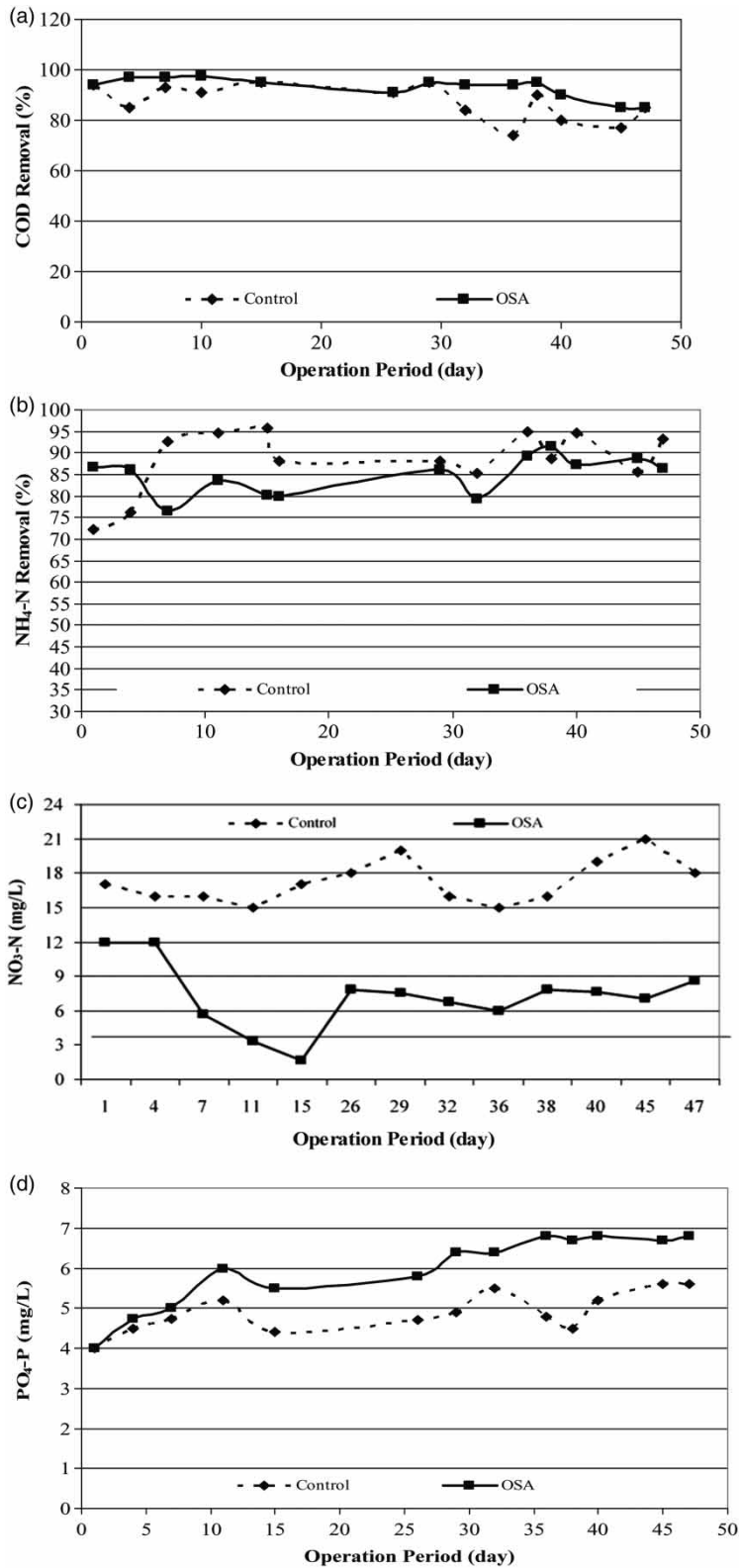


Figure 2 | Changes of (a)  $Y_{obs}$ , (b) MLSS and (c) MLVSS in OSA and control system.

### Comparison of OSA and control system in terms of effluent quality

Figure 3(a) summarizes the COD removal efficiencies of the systems. It was found that COD concentrations in the effluent

in the OSA process were lower than in the effluent of the control system due to the additional substrates from the anaerobic tank. The results indicated that when compared with the control reactor, the reactor that was fed with sludge from the anaerobic tank showed good removal



**Figure 3** | (a) Changes of COD removal efficiencies (%) in OSA and control system. (b) Changes of  $\text{NH}_4\text{-N}$  removal efficiencies (%) in OSA and control system. (c) Changes of  $\text{NO}_3\text{-N}$  concentration of effluent in OSA and control system. (d) Changes of  $\text{PO}_4\text{-P}$  concentration of effluent in OSA and control system.



efficiency for COD. The high removal efficiencies in the OSA process mean that the insertion of the anaerobic tank actually improves COD removal under the same influent COD concentration. It can be explained that when sludge is exposed to low ORP level in the anaerobic tank, the sludge may also be subject to starvation conditions which promote substrate removal ability in the following aeration tank with the presence of food (Chen *et al.* 2001; Saby *et al.* 2003). The  $\text{NH}_4\text{-N}$  removal efficiency was lower compared to the control system during the operation period due to denitrification. Figure 3(b) shows the  $\text{NH}_4\text{-N}$  removal capacity of the OSA and control systems. In the control system, the removal of  $\text{NH}_4\text{-N}$  increased and reached 96%, whereas in the OSA process, the removal of  $\text{NH}_4\text{-N}$  decreased from 87 to 76% due to the effect of denitrification. However, in long-term operation, it can be said that both systems showed similar removal efficiencies for  $\text{NH}_4\text{-N}$ .

As seen in Figure 3(c), in the OSA process, the  $\text{NO}_3\text{-N}$  concentration of effluent decreased significantly and it can

be attributed to the denitrification as was also seen in the study by Saby *et al.* (2003).

Phosphorus release after the anaerobic conditions was proved with increasing of the  $\text{PO}_4\text{-P}$  concentration of effluent in the OSA process as shown in Figure 3(d). The  $\text{PO}_4\text{-P}$  concentration of effluent in the OSA process was increased from 4 to 6.8 mg/L due to the addition of sludge kept under  $-250$  mV ORP level with no external food.

The presence of the anaerobic reactor also affects the release of phosphorus; a higher increase of  $\text{PO}_4$  concentration in the treated effluent from the wastewater treatment units was observed for lower ORP values.

### Effects of anaerobic zone of OSA on TN and TP concentrations

TP concentration in the aeration tank in the OSA process was gradually increased from 3 to 8.6 mg/L due to the release of phosphorus in the anaerobic tank as seen in Figure 4(a).

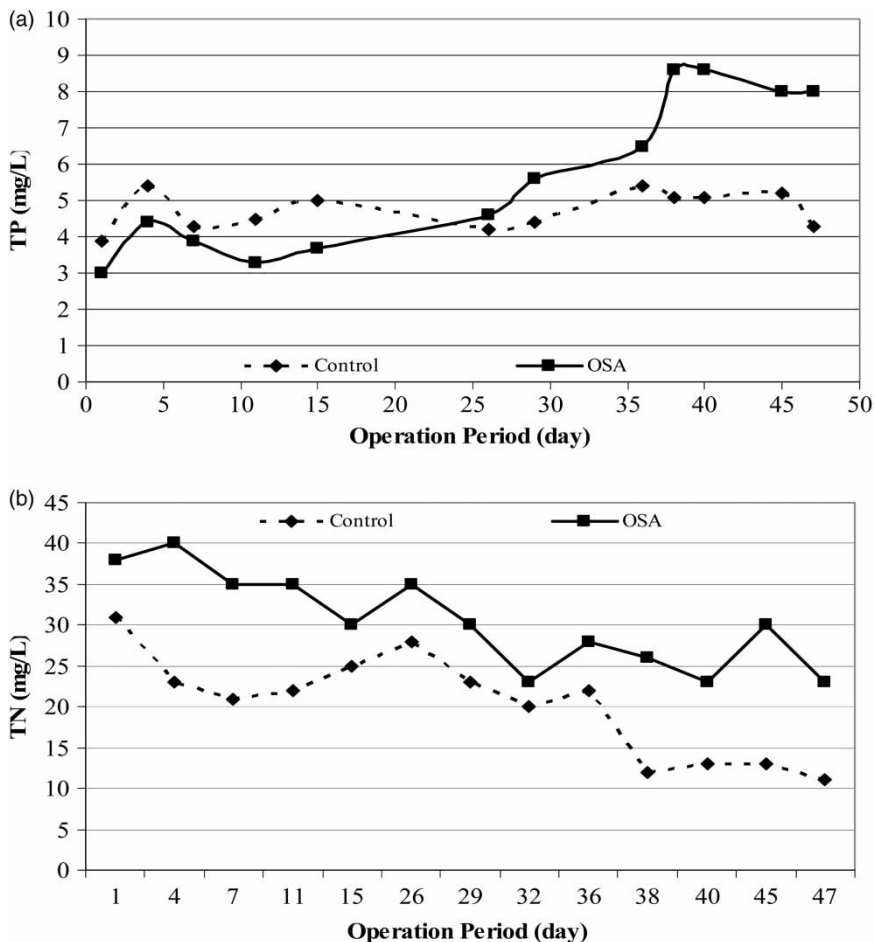


Figure 4 | (a) Changes of TP concentration of aeration tank in OSA and control system. (b) Changes of TN concentration of aeration tank in OSA and control system.

Previous studies have concluded that there are two common aspects related to the volatile suspended solids (VSS) reduction in conventional aerobic digestion and in alternate aerobic/anoxic phases. One of them is the similarity of the VSS reduction in the alternating aerobic/anoxic phases to that obtained in the conventional aerobic digestion. The other one is the significant increase of the total N reduction due to the introduction of denitrification during the anoxic phase (Foladori *et al.* 2010). The second aspect was proved with Figure 4(b). TN concentration of the OSA process was higher than the control system as shown in Figure 4(b) due to the denitrification and release of cell contents after the anaerobic tank. The adoption of aerobic anoxic phases during digestion leads to good total N removal efficiency with lower nitrogen loads recirculated to the wastewater handling units (Foladori *et al.* 2010).

### Comparison of OSA and control system in terms of sludge characteristics

The settleability of the OSA process sludge was also found to be better than that of the control system in terms of SVI as shown in Figure 5(a). Over the entire operation, average SVI values were 94 and 115 mL/gMLSS for the OSA and control systems, respectively.

Similar to this study, Saby *et al.* (2003) suggested an improvement in settleability in OSA sludge with around 100 mL/gMLSS of SVI values. The maintenance of low SVI was attributed to the low ORP level in the anaerobic tank. It can be revealed that low ORP level was necessary to maintain low SVI values.

During continuous operation of the OSA and control systems, CST parameter was used for the evaluation of filtration and the dewatering characteristics of sludge. CST variations during the operation period are given in Figure 5(b). In the OSA process, CST values were higher than the control system at the beginning of the operation. With increasing operating time, the CST values decreased in the OSA process. Higher CST values were obtained from the control system compared to the OSA process during the long-term operation. This shows that the OSA process enhanced the filterability of sludge.

SRF is a relatively complicated method compared to CST and gives information about sludge behavior on vacuum filtration units. Similar to the CST parameter, low SRF values indicate a good dewatering characteristic of sludge. The OSA process improved the filterability characteristics of sludge in long-term operation in terms of low SRF values as shown in Figure 5(c) and it can be applied

to biological sludge without deterioration of sludge filterability.

The reduction in particle size generally allows an easier hydrolysis of solids within the sludge due to the larger surface areas in relation to the particle volumes. The result is an accelerated and enhanced degradation of the organic fraction of the solid phase (Xie *et al.* 2009). The particle size distribution in the control and OSA process are illustrated in Figure 5(d) and 5(e), respectively. These figures show the percentage of particles, by volume. A different size distribution was obtained for each system. The sludge from the control and the OSA process showed peaks in the size range 10–100 µm. There was a shift in the particle size distribution towards larger particles with the peak at 1,000 µm. It can be attributed to the differences in biodegradability of the sludge. Larger particles, more resistant to degradation, are therefore likely to be the cause of the shift in the particle size distribution.

### Comparison of OSA and control system in terms of OUR and SOUR

The profile of OUR is valuable for determining the solubilization of activated sludge. Monitoring of OUR value is an effective technique for investigating the performance in aerobic, anoxic and anaerobic conditions. The increase in OUR value leads to the increase in SCOD concentration (Chang *et al.* 2002). At the beginning of the operation period, OUR values in the OSA process increased due to the COD solubilization. After the 15th day of the period, OUR values decreased to 0.15 mg/L.min as shown in Figure 6(a).

SOUR values in the OSA process were also increased due to OUR values as illustrated in Figure 6(b). The milligram of oxygen consumed per gram of VSS per hour was increased after the anaerobic tank under low ORP level during the operational period.

### Batch experiments of OSA process

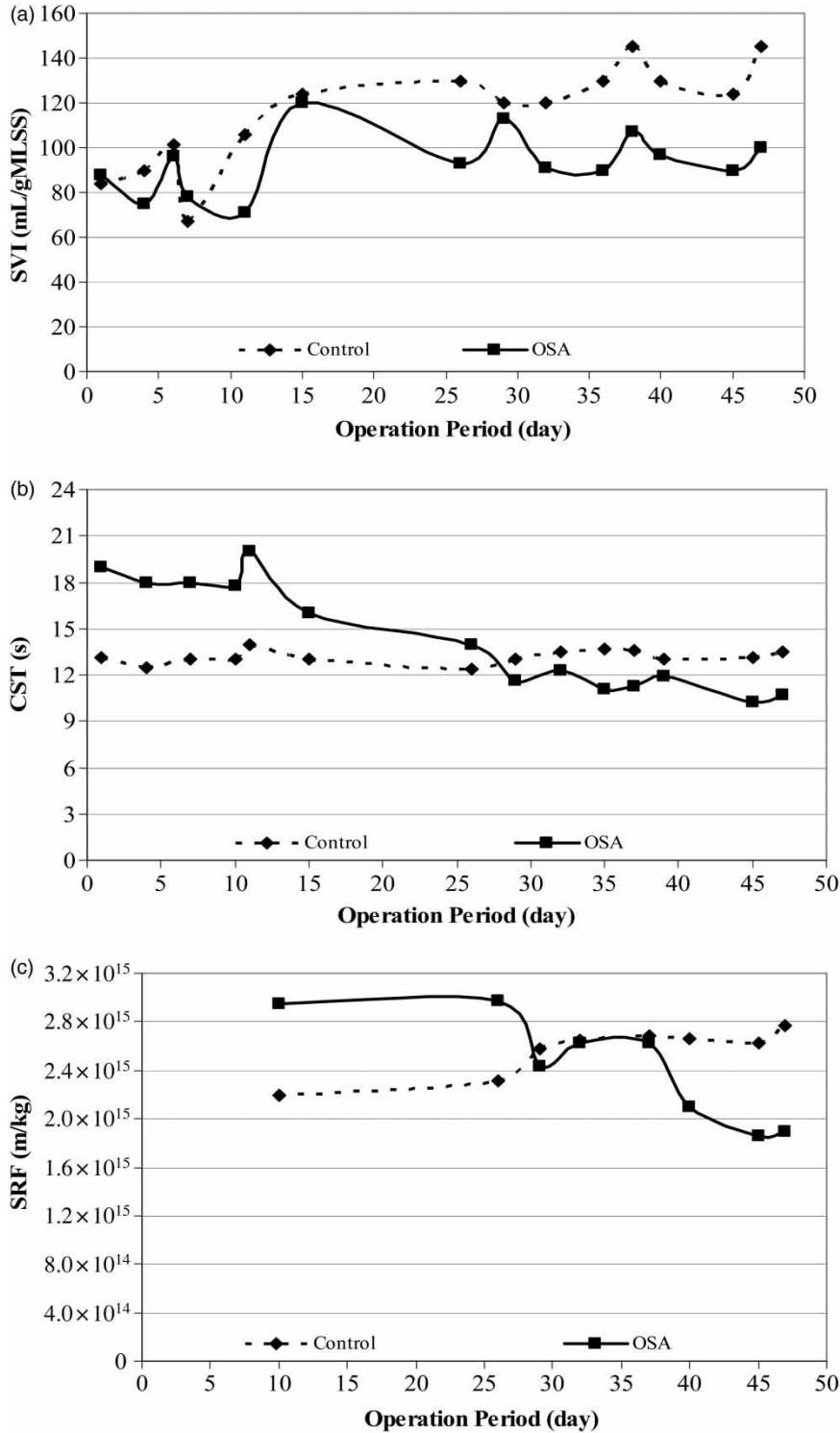
#### Batch experiment I: sludge decay

A possible explanation of sludge reduction is that the rate of cell death accelerated when sludge is subjected to low ORP values in the anaerobic reactor. In the anaerobic reactor, an increase of soluble COD was observed. The solubilized COD could be related to cellular death. According to this hypothesis, the anaerobic reactor behaves like a cell lysis reactor and when the sludge returns to the aerobic activated



sludge stage, the soluble COD promotes cryptic growth, which leads to an overall sludge reduction. The effect of the 18 h anaerobic conditions of the MLSS concentration

is demonstrated in Figure 7(a). MLSS concentration was reduced by 19% in the control system after the anaerobic stage while 13% of MLSS reduction was observed in the



**Figure 5** | (a) Changes of SVI values in OSA and control system. (b) Changes of CST values in OSA and control system. (c) Changes of SRF values in OSA and control system. (d) Changes of particle size in control system. (e) Changes of particle size in OSA process. (Continued.)

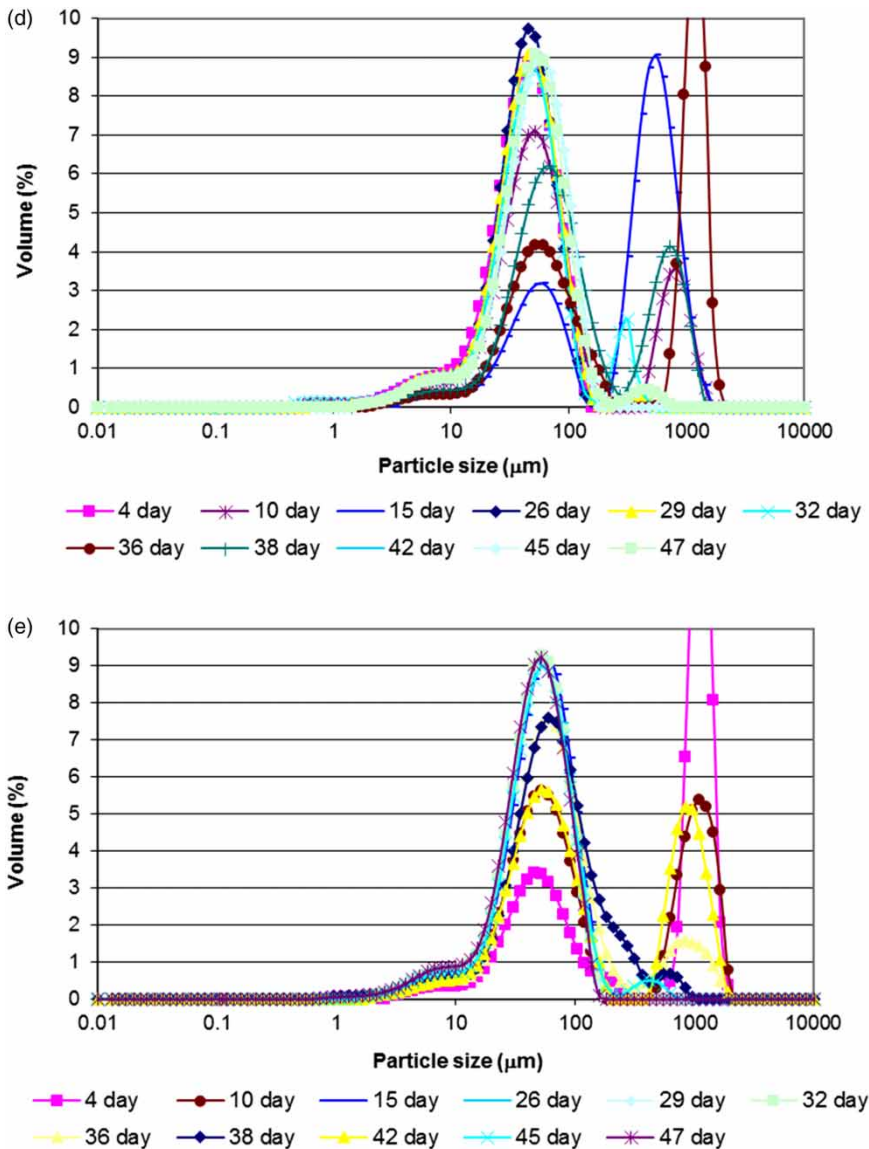


Figure 5 | Continued.

sludge from the OSA process during 18 h. Low ORP level and absence of external food may promote sludge reduction by the stress they place upon the microorganisms. The stress on the OSA process was lower than on the control system because microorganisms have already acclimated to the low ORP level.

As shown in Figure 7(b) and 7(c) during the anaerobic exposure, SCOD,  $\text{NH}_4\text{-N}$ , TN and TP in the supernatant were increased. At the end of the anaerobic treatment, the released cell contents may contribute to the increase of SCOD,  $\text{NH}_4\text{-N}$ , TN and TP. The concentration of COD and nitrogen components were low because of the anaerobic exposure. As a result of the anaerobic treatments of sludge

with no food, the hydrolysis and acidogenesis processes occurred. In the hydrolysis phase, organic compounds were hydrolyzed to simple components. Then simple organics were degraded to volatile fatty acids and the sludge decay led to the reduction of excess sludge production.

The changes of OUR and SOUR before and after anaerobic treatment are shown in Figure 7(d). These values for the control system were lower than for the OSA process before anaerobic treatment. The 18 h anaerobic period affected them for both the control system and OSA process. The increase of OUR and SOUR may be an indicator of the endogenous metabolism. After anaerobic treatment, the cellular components were oxidized to

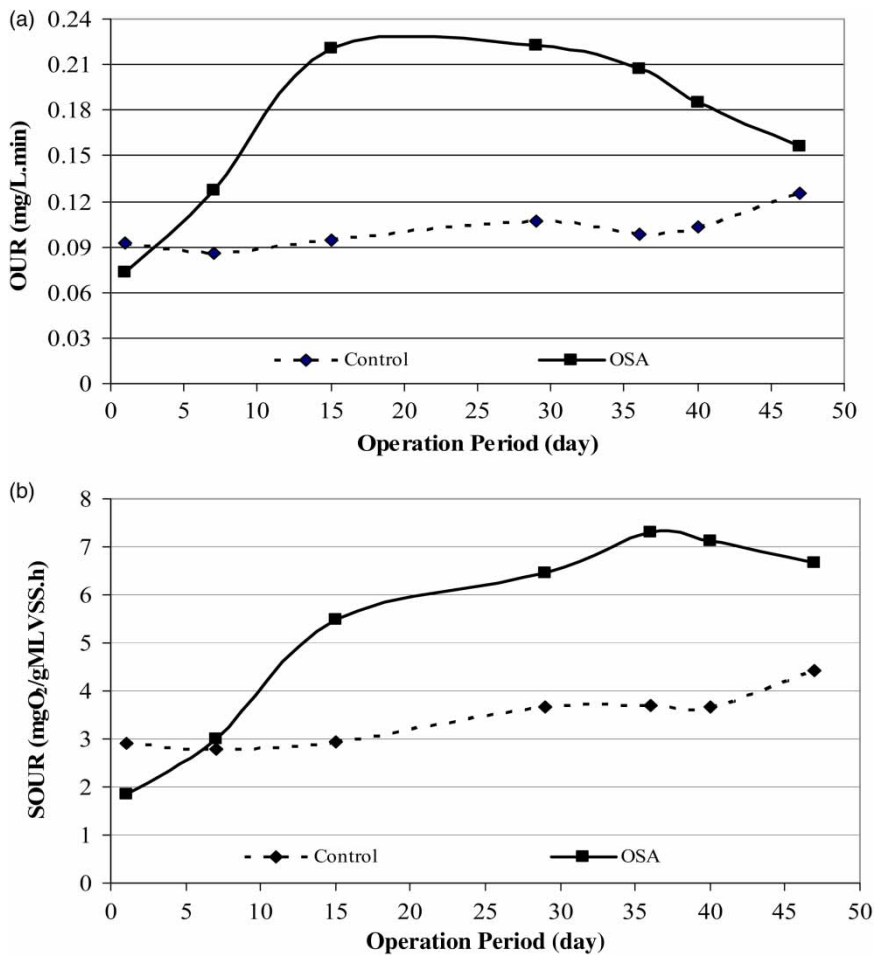


Figure 6 | (a) Changes of OUR values in OSA and control system. (b) Changes of SOUR values in OSA and control system.

produce maintenance energy and there was no energy for growth. Therefore, the sludge production was decreased after anaerobic treatment for both the control and the OSA process as demonstrated before by MLSS reduction. This shows that higher endogenous respiration in the systems might lead to lower sludge production.

$Y_{obs}$  values in the batch OSA and control systems are presented in Table 3. The sludge reduction in terms of  $Y_{obs}$  caused by sludge decay in the OSA batch reactor was 40% compared to the control batch reactor for the first batch experiment.

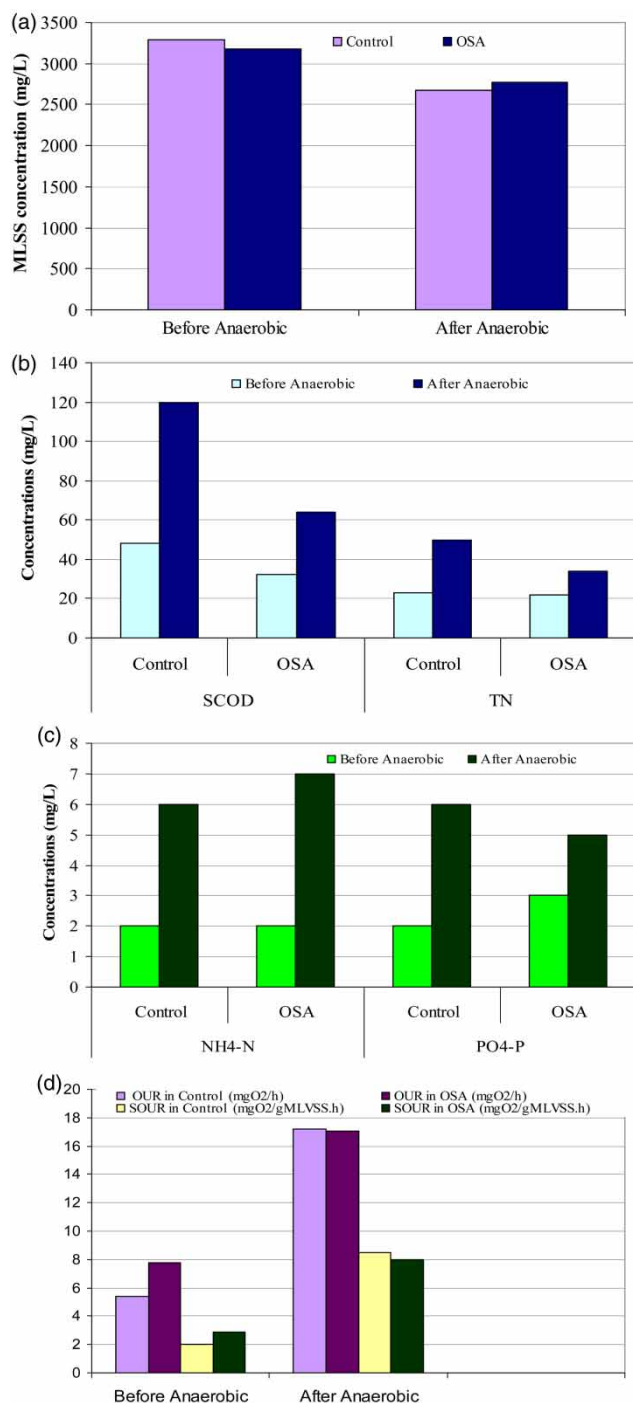
### Batch experiment II: energy uncoupling

A decrease of ATP content in sludge occurs when the sludge remains in the anaerobic reactor, without external substrate and at low ORP levels. When the sludge returns to the activated sludge stage, optimal conditions for the formation of

new ATP are stored. The cyclic alternation of ATP content in sludge uncouples catabolism and anabolism, which causes decrease in biomass yield, favoring sludge reduction.

Results in Table 4 show that there was an energy uncoupling for the sludge reduction in the anaerobic-aerobic batch experiment.

The sludge reduction in terms of  $Y_{obs}$  caused by energy uncoupling in the OSA batch reactor was 5% compared to the control batch reactor for the second batch experiment. The first study to evaluate the reduction of excess sludge by the OSA process was carried out by Chudoba *et al.* (1992). At laboratory scale and using synthetic wastewater, the sludge production of a CAS system was compared to that of the OSA process. In the OSA process, the anaerobic reactor was maintained at  $-250$  mV ORP, similar to this study. The observed sludge yield in the activated sludge was  $0.37$  kgTSS/kgCOD<sub>removed</sub>, while in the OSA process, it was  $0.22$  kgTSS/kgCOD<sub>removed</sub> (a reduction of 40%). Saby *et al.*



**Figure 7** | (a) MLSS concentrations of control and OSA batch experiment. (b) SCOD and TN concentrations of control and OSA batch experiment. (c) NH<sub>4</sub>-N and TP concentrations of control and OSA batch experiment. (d) Change of OUR and SOUR after anaerobic treatment in control and OSA.

(2003) also reported that the observed sludge yields in the OSA process were 0.18–0.32 kgTSS/kgCOD<sub>removed</sub> compared to 0.4 kgTSS/kgCOD<sub>removed</sub> in the conventional activated sludge process.

**Table 3** | Y<sub>obs</sub> in the batch experiment I

Measurements	Control batch reactor	OSA batch reactor
1	0.42	0.25
2	0.39	0.27
3	0.41	0.24
Average	0.41	0.25

**Table 4** | Y<sub>obs</sub> in the batch experiment II

Measurements	Control batch reactor	OSA batch reactor
1	0.4	0.39
2	0.39	0.38
3	0.41	0.37
Average	0.4	0.38

## CONCLUSION

In this study, 58% yield reduction efficiency was achieved in the OSA process compared to the control system at the end of the operation. The settleability of the OSA process sludge was also found to be better than that of the control system in terms of SVI. The OSA process also enhanced the filterability of sludge. It was shown that higher endogenous respiration in the systems might lead to lower sludge production. There was a little energy uncoupling effect of the sludge reduction. This study revealed that the OSA process can be used as a sludge minimization technique with good effluent quality and sludge characteristics.

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